ever, the sub-unit vaccines use adjuvant for enhancing immune responses since the vaccines show lower immunogenicity compared to the prior art vaccines.

[0012] Since antibodies act as primary defense actors against most of pathogenic bacteria or viruses, only antibodies induced by vaccine antigens can prevent various diseases. But, since cell-mediated immune responses act significantly on infection diseases against which vaccines have not been developed in preventions or treatments. In this case, it is possible to develop vaccines efficiently when using adjuvant inducing cell-mediated immune response.

[0013] Currently, alum, metal salts such as aluminum hydroxide, aluminum phosphate or aluminum hydroxide phosphate sulphate, and MF59, oil-in-water emulsion type adjuvant based on squalene, have been mainly used as adjuvants for human vaccines adjuvant. Such commonly used adjuvants induce little cell-mediated immunity while induce mainly humoral immunity. Accordingly, such adjuvants can be utilized only in case antibodies can defend infections, and they were not proper for vaccines requiring cell-mediated immune responses.

[0014] Micro-organisms as the typical pathogens have pathogen-associated molecular patterns (PAMPs) such as lipopolysaccharide (LPS), betha-1,3-glucan and peptidoglycans in cell walls thereof. A specific protein consisting of immune system of a host, for example, pattern recognition receptors (PRRs) or pattern recognition proteins (PRPs) can recognize such PAPMs. Each of PRRs or PRPs can recognize a proper PAMPs on the surface of the pathogens to form a complex that induce a series of immune responses such phagocytosis, nodule formation, encapsulation, proteinase cascade activation, and anti-bacterial peptides synthesis. Toll-like receptors (TLRs) are representative PRR, and TLR agonist have been developed as vaccine adjuvants because they show strong activities to immunocytes. For example, an endotoxin LPS showed strong immunity activities against TLR4 on immunocytes.

[0015] Unlike genomic DNA in higher organism such as human, bacterial DNA does not have methylated cytosine in CpG motif. The immunocytes in higher organisms can bacterial DNA in which cytosine of CpG motif is not methylated as non-self antigens. In this case, a specific receptor TLR9 recognizes the bacterial DNA. TLR9 agonists can enhance various immune responses, and TLR9 agonist such as oligo-nucleotides including CpG motif have been developed as adjuvants. However, LPS and CpG motif used as TLR agonists have very strong toxicity, causes cases side effects such as inflammatory response in the body.

DISCLOSURE

Technical Problem

[0016] Accordingly, the present disclosure is directed to a nucleic acid molecule, an expression vector and pharmaceutical or medicinal applications that can reduce one or more of the problems due to the limitations and disadvantages of the related art.

[0017] An object of the present disclosure is to provide an expression system that express peptides or proteins of interest efficiently without incurring complex and expensive processes.

[0018] Another object of the present disclosure is to provide a pharmaceutical composition such as adjuvant that can induce or stimulate cell-mediated immune response as well as humoral immune response.

Solution to Problem

[0019] According to an aspect, the present disclosure provides a nucleic acid molecule comprises at least one expression control sequence comprising a viral Internal Ribosomal Entry Site (IRES) element; and at least one coding region linked operatively to the at least one expression control sequence and encoding a peptide or a protein.

[0020] In one embodiment, the nucleic acid molecule may further comprise at least one of multiple adenosines and multiple thymidines located upstream of the at least one expression control sequence.

[0021] The viral IRES element may be derived from at least one of Picornaviridae family, Togaviridae family, Dicistroviridae family, Flaviridae family, Retroviridae family and Herpesviridae family, for example, may be derived from at least one of Picornaviridae family and Dicistroviridae family.

[0022] In an exemplary embodiment, the viral IRES element derived from the Picornaviridae may be derived from at least one of Enterovirus genus, Cardiovirus genus, Aphthovirus genus, Hepatovirus genus and Teschovirus genus, and the viral IRES element derived from the Dicistroviridae family may be derived from Cripavirus genus. For example, the viral IRES element may be derived from at least one of coxsackie B virus, Cricket paralysis virus, Japanese Encephalitis virus, Encephalomyocarditis virus and Sindbis virus.

[0023] In another exemplary embodiment, the at least one expression control sequence may comprise a viral 5' untranslated region (5' UTR). If necessary, the nucleic acid molecule may further comprise a viral 3' Untranslated Region (3' UTR) located downstream of the 5' UTR, and wherein the at least one coding region is located between the 5' UTR and the 3' UTR.

[0024] In one embodiment, the at least one coding region may encode an antigen or fragments thereof. Alternatively, the at least one coding region encodes a protein or fragments thereof for treating disease.

[0025] In another exemplary embodiment, the at least one expression control sequence comprises a first expression control sequence having a first IRES element and a second expression control sequence located downstream of the first expression control sequence and having a second IRES element. The at least one coding region may comprise a first coding region located between the first and second expression control sequences and a second coding region located downstream of the second expression control sequence. The nucleic acid molecule may further comprise at least one of multiple adenosines or multiple thymidines located upstream of at least one of the first expression control sequence and the second expression control sequence. The first expression control sequence may comprise a first viral IRES element derived from coxsackie B virus or Cricket paralysis virus, and the second expression control sequence may comprise a second viral IRES element derived from Encephalomyocarditis virus.

[0026] Alternatively, the nucleic acid molecule may further comprise a transcription control sequence located downstream of the at least one expression control sequence, and/or a polyadenylation signal sequence or a poly adenos-